OSMOLYTE EFFLUX DURING REGULATED VOLUME DECREASE IN *SALMO* <u>SALAR</u> ERYTHROCYTES, L.Z. <u>Mason</u>, D.B. Light*, Department of Biology, Lake Forest College, Lake Forest, IL 60045, <u>light@lakeforest.edu</u>

The ability of cells to regulate their volume is critical for the maintenance of homeostasis. The aim of this study was to assess the efflux pathways of regulated volume decrease (RVD) in Atlantic salmon (Salmo salar) red blood cells. The volume of cells placed in various Ringer solutions was measured electronically using a Z2 Coulter counter. Hypotonic (0.5X) NaCl (67 - 89mM), KCl (67 - 90mM), and taurine (45 -100mM) Ringer solutions were used to identify which osmolytes were lost following cell swelling. Cells placed in a Ringer in which potassium had been substituted for sodium showed an accelerated recovery for the first 20 minutes; recovery was then inhibited over the remainder of observation (90 min). In contrast, bathing cells with an elevated taurine Ringer inhibited RVD throughout (90 min), suggesting that removing the driving force for taurine efflux limits volume recovery. To elucidate the steps that result in osmolyte efflux, experiments were modeled after studies using red cells from an aquatic salamander (Necturus maculosus: Light et al. 2001. J. Membr. Biol. 182: 193), in which the efflux and autocrine binding of ATP to a purinergic receptor (subclass P2) was an initial step in RVD. However, in Atlantic salmon, the addition of extracellular ATP (100 μM) or hexokinase (2.5 U/mL), an enzyme that degrades ATP in the presence of glucose, had no effect. Taken together, these results suggest that Salmo erythrocytes loose a combination of taurine and potassium during RVD, and the P2 class of purinoreceptors are not involved in this process. Future studies will use an impermeant cation to assess the possibility of initial sodium uptake during RVD, which could explain an initial accelerated recovery in a hypotonic high K⁺ Ringer. Additionally, quinine, a potassium channel blocker, and DIDS, which blocks chloride transport, will be used to further evaluate the nature of osmolyte efflux. Adenosine will also be used to determine whether P1 receptors may be present.

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